specific receptors). Preferred complexes are sufficiently stable *in vivo* to prevent significant uncoupling prior to internalization by the target cell. However, the complex is cleavable under appropriate conditions within the cell so that the gene, protein, polypeptide or peptide is released in a functional form. It is also possible that soluble forms of the protein also exist. Such soluble isoforms can arise through variable splicing of the HKNG1 gene or alternatively as a result of proteolysis of a membranous isoform.

Please replace the paragraph beginning at page 11, line 10 with the following rewritten paragraph:

As is well known, genes for a particular polypeptide may exist in single or multiple copies within the genome of an individual. Such duplicate genes may be identical or may have certain modifications, including nucleotide substitutions, additions or deletions, which all still code for polypeptides having substantially the same activity. The term "DNA sequence encoding an HKNG1 polypeptide" may thus refer to one or more genes within a particular individual. Moreover, certain differences in nucleotide sequences may exist between individual organisms, which are called alleles. Such allelic differences may or may not result in differences in amino acid sequence of the encoded polypeptide yet still encode a protein with the same biological activity.

Please replace the paragraph beginning at page 11, line 19 with the following rewritten paragraph:

As used herein, the term "gene" or "recombinant gene", as applied to *HKNG1*, refers to a polynucleotide or nucleic acid molecule comprising an open reading frame encoding one of the HKNG1 polypeptides of the present invention. In one embodiment, these terms relate to a cDNA sequence including, but not limited to, a polynucleotide or nucleic acid sequence obtained via reverse transcription of an mRNA molecule. In one embodiment, the term nucleic acid or polynucleotide is a nucleic acid molecule which is not genomic but is a cDNA derived from a contiguous coding region which includes, but is not limited to, reverse transcribed cDNA. In another embodiment, the term nucleic acid or polynucleotide refers to a nucleic acid molecule which comprises contiguous nucleotide codons. In yet another embodiment, the term nucleic

C1 Cont'

C1 cont'd

acid or polynucleotide is a nucleic acid molecule which is genomic but which excludes intronic sequences.

Please replace the paragraph beginning at page 12, line 10 with the following rewritten paragraph:

C2

Furthermore, a degree of homology or similarity of amino acid sequences is a function of the number of conserved amino acids at positions shared by the amino acid sequences. A sequence which is "unrelated" or "non-homologous" with one of the human HKNG1 sequences of the present invention typically is a sequence which shares less than 40 % identity, though preferably less than 25 % identity with one of the human HKNG1 sequences of the present invention.

Please replace the paragraph beginning at page 13, line 21 with the following rewritten paragraph:

C3

As used herein, the term "hybridizes under stringent conditions" is intended to describe conditions for hybridization and washing under which nucleotide sequences at least 60% (65%, 70%, preferably 75% or more) identical to each other typically remain hybridized to each other. Such stringent conditions are known to those skilled in the art and can be found in *Current Protocols in Molecular Biology*, John Wiley & Sons, N.Y. (1989), 6.3.1-6.3.6. A preferred, non-limiting example of stringent hybridization conditions are hybridization in 6X sodium chloride/sodium citrate (SSC) at about 45°C, followed by one or more washes in 2.0 X SSC at 50° C. (low stringency) or 0.2 X SSC, 0.1% SDS at 50-65°C (high stringency). In one embodiment, an isolated nucleic acid molecule of the invention that hybridizes under stringent conditions to the sequence of SEQ ID NOS:1, 3, 5, 6, or 7, or to a complement thereof, corresponds to a naturally-occurring nucleic acid molecule.

Please replace the paragraph beginning at page 14, line 23 with the following rewritten paragraph:

C4

FIG. 3A-3II. Genomic sequences of the human *HKNG1* gene (SEQ ID NO:7). Exons are in bold and the 3' and 5' UTR's are underlined.

Please replace the paragraph beginning at page 14, line 25 with the following rewritten paragraph:

C4 cont

FIG. 4A-4B. Summary of *in situ* hybridization analysis of *HKNG1* mRNA distribution in normal human brain tissue.

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Please replace the paragraph beginning at page 40, line 5 with the following rewritten paragraph:

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C5

Once the sequence (or a portion of the sequence) of a gene has been isolated, this sequence can be used to map the location of the gene on a chromosome. Accordingly, nucleic acid molecules described herein or fragments thereof, can be used to map the location of the corresponding genes on a chromosome. For example, the nucleic acid molecules described herein can be used to map the chromosomal location of *HKNG1* homologues in various species. Such mapping information can be used, for example, for analysis of the activity of *HKNG1* transgenes in mice. The nucleic acid molecules can further be used to map the location of copies of *HKNG1* genes in the human chromosome, such as those caused by genetic abnormalties, e.g., translocations.

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Please replace the paragraph beginning at page 41, line 10 with the following rewritten paragraph:

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C6

In another embodiment, a HKNG1 polypeptide and fragments and sequences thereof and antibodies specific thereto can be used to map the location of the gene encoding the polypeptide on a chromosome. This mapping can be carried out by specifically detecting the presence of the polypeptide in members of a panel of somatic cell hybrids between cells of a first species of animal from which the protein originates and cells from a second species of animal and then determining which somatic cell hybrid(s) expresses the polypeptide and noting the chromosome(s) from the first species of animal that it contains. For examples of this technique, see Pajunen *et al.* (1988) *Cytogenet. Cell Genet. 47*:37-41 and Van Keuren *et al.* (1986) *Hum. Genet. 74*:34-40. Alternatively, the presence of the polypeptide in the somatic cell hybrids can be determined by assaying an activity or property of the polypeptide, for example, enzymatic

activity, as described in Bordelon-Riser *et al.* (1979) *Somatic Cell Genetics 5*:597-613 and Owerbach *et al.* (1978) *Proc. Natl. Acad. Sci. USA 75*:5640-5644.

Please replace the heading beginning at page 44, line 13 with the following rewritten heading:

USE OF *HKNG1* GENE SEQUENCES IN PREDICTIVE MEDICINE

Please replace the paragraph beginning at page 44, line 15 with the following rewritten paragraph:

The present invention also pertains to the field of predictive medicine in which diagnostic assays, prognostic assays, and monitoring clinical trials are used for prognostic (predictive) purposes to thereby treat an individual prophylactically. Accordingly, one aspect of the present invention relates to diagnostic assays for determining HKNG1 protein and/or nucleic acid expression as well as HKNG1 activity, in the context of a biological sample (e.g., blood, serum, cells, tissue) to thereby determine whether an individual is afflicted with a disease or disorder, or is at risk of developing a disorder, associated with aberrant or unwanted HKNG1 expression or activity. The invention also provides for prognostic (or predictive) assays for determining whether an individual is at risk of developing a disorder associated with an HKNG1 protein, nucleic acid expression or activity. For example, mutations in an HKNG1 gene can be assayed in a biological sample. Such assays can be used for prognostic or predictive purpose to thereby prophylactically treat an individual prior to the onset of a disorder characterized by or associated with an HKNG1 protein, nucleic acid expression or activity.

Please replace the paragraph beginning at page 44, line 29 with the following rewritten paragraph:

As an alternative to making determinations based on the absolute expression level of selected genes, determinations may be based on the normalized expression levels of these genes. Expression levels are normalized by correcting the absolute expression level of an *HKNG1* gene by comparing its expression to the expression of a gene that is not an *HKNG1* gene, e.g., a housekeeping gene that is constitutively expressed. Suitable genes for normalization include

housekeeping genes such as the actin gene. This normalization allows the comparison of the expression level in one sample, e.g., a patient sample, to another sample, e.g., a non-BAD-affected normal sample, or between samples from different sources.

Please replace the paragraph beginning at page 45, line 6 with the following rewritten paragraph:

Alternatively, the expression level can be provided as a relative expression level. To determine a relative expression level of a gene, the level of expression of the gene is determined for 10 or more samples of different cell isolates, preferably 50 or more samples, prior to the determination of the expression level for the sample in question. The cell isolates are selected depending upon the tissues in which the gene of interest is expressed. For example, for HKNG1 family members, expression was observed in the brain. The mean expression level of each of the genes assayed in the larger number of samples is determined and this is used as a baseline expression level for the gene(s) in question. The expression level of the gene determined for the test sample (absolute level of expression) is then divided by the mean expression value obtained for that gene. This provides a relative expression level and aids in identifying extreme cases of a HKNG1-mediated disease.

Please replace the paragraph beginning at page 45, line 18 with the following rewritten paragraph:

For example, by way of illustration only, for HKNG1 family members, diseases which may be studied include, without limitation, those associated with tissues of the brain.

Please replace the paragraph beginning at page 45, line 21 with the following rewritten paragraph:

Preferably, the samples used in the baseline determination will be from an HKNG1-mediated diseased or from non-diseased cells of tissue. The choice of the cell source is dependent on the use of the relative expression level. Using expression found in normal tissues as a mean expression score aids in validating whether the *HKNG1* gene assayed is cell-type specific for the tissues in which expression is observed versus the expression found in normal

C7 cont

cells. Such a use is particularly important in identifying whether an *HKNG1* gene can serve as a target gene. In addition, as more data is accumulated, the mean expression value can be revised, providing improved relative expression values based on accumulated data. Expression data from brain cells provides a means for grading the severity of the HKNG1-mediated disease state.

Please replace the paragraph beginning at page 45, line 31 with the following rewritten paragraph:

C7 cont'd

Another aspect of the invention pertains to monitoring the influence of agents (e.g., drugs, compounds) on the expression or activity of HKNG1 in clinical trials.

Please replace the paragraph beginning at page 52, line 3 with the following rewritten paragraph:

C8

Accordingly, in one aspect, the invention provides substantially purified antibodies or fragment thereof, and non-human antibodies or fragments thereof, which antibodies or fragments specifically bind to a polypeptide comprising an amino acid sequence selected from the group consisting of: the amino acid sequence of any one of SEQ ID NOS:2 or 4; a fragment of at least 15 amino acid residues of the amino acid sequence of any one of SEQ ID NOS:2 or 4, an amino acid sequence which is at least 95% identical to the amino acid sequence of any one of SEQ ID NOS:2 or 4, wherein the percent identity is determined using the ALIGN program of the GCG software package with a PAM120 weight residue table, a gap length penalty of 12, and a gap penalty of 4; and an amino acid sequence which is encoded by a nucleic acid molecule which hybridizes to the nucleic acid molecule consisting of any one of SEQ ID NOS:1, 3, 5, 6, or 7, or a complement thereof, under conditions of hybridization of 6X SSC at 45°C and washing in 0.2 X SSC, 0.1% SDS at 65°C. In various embodiments, the substantially purified antibodies of the invention, or fragments thereof, can be human, non-human, chimeric and/or humanized antibodies.

Please replace the paragraph beginning at page 52, line 18 with the following rewritten paragraph:

In another aspect, the invention provides non-human antibodies or fragments thereof, which antibodies or fragments specifically bind to a polypeptide comprising an amino acid sequence selected from the group consisting of: the amino acid sequence of any one of SEQ ID NOS:2 or 4; a fragment of at least 15 amino acid residues of the amino acid sequence of any one of SEQ ID NOS:2 or 4, an amino acid sequence which is at least 95% identical to the amino acid sequence of any one of SEQ ID NOS:2 or 4, wherein the percent identity is determined using the ALIGN program of the GCG software package with a PAM120 weight residue table, a gap length penalty of 12, and a gap penalty of 4; and an amino acid sequence which is encoded by a nucleic acid molecule which hybridizes to the nucleic acid molecule consisting of any one of SEQ ID NOS:1, 3, 5, 6, or 7, or a complement thereof, under conditions of hybridization of 6X SSC at 45°C and washing in 0.2 X SSC, 0.1% SDS at 65°C. Such non-human antibodies can be goat, mouse, sheep, horse, chicken, rabbit, or rat antibodies. Alternatively, the non-human antibodies of the invention can be chimeric and/or humanized antibodies. In addition, the non-human antibodies of the invention can be polyclonal antibodies or monoclonal antibodies.

Please replace the paragraph beginning at page 53, line 1 with the following rewritten paragraph:

In still a further aspect, the invention provides monoclonal antibodies or fragments thereof, which antibodies or fragments specifically bind to a polypeptide comprising an amino acid sequence selected from the group consisting of: the amino acid sequence of any one of SEQ ID NOS:2 or 4; a fragment of at least 15 amino acid residues of the amino acid sequence of any one of SEQ ID NOS:2 or 4, an amino acid sequence which is at least 95% identical to the amino acid sequence of any one of SEQ ID NOS:2 or 4, wherein the percent identity is determined using the ALIGN program of the GCG software package with a PAM120 weight residue table, a gap length penalty of 12, and a gap penalty of 4; and an amino acid sequence which is encoded by a nucleic acid molecule which hybridizes to the nucleic acid molecule consisting of any one of SEQ ID NOS:1, 3, 5, 6, or 7, or a complement thereof, under conditions of hybridization of 6X SSC at 45°C and washing in 0.2 X SSC, 0.1% SDS at 65°C. The monoclonal antibodies can be human, humanized, chimeric and/or non-human antibodies.

C8 cont

Please replace the paragraph beginning at page 53, line 15 with the following rewritten paragraph:

The substantially purified antibodies or fragments thereof specifically bind to a signal peptide, a secreted sequence, an extracellular domain, a transmembrane or a cytoplasmic domain cytoplasmic membrane of a polypeptide of the invention. In one embodiment, the substantially purified antibodies or fragments thereof, the human or non-human antibodies or fragments thereof, and/or the monoclonal antibodies or fragments thereof, of the invention specifically bind to a secreted sequence or an extracellular domain of the amino acid sequence of SEQ ID NOS:2 or 4.

Please replace the paragraph beginning at page 54, line 1 with the following rewritten paragraph:

Still another aspect of the invention is a method of making an antibody that specifically recognizes HKNG1, the method comprising immunizing a mammal with a polypeptide. The polypeptide used as an immungen comprises an amino acid sequence selected from the group consisting of: the amino acid sequence of any one of SEQ ID NOS:2 or 4; a fragment of at least 15 amino acid residues of the amino acid sequence of any one of SEQ ID NOS:2 or 4, an amino acid sequence which is at least 95% identical to the amino acid sequence of any one of SEQ ID NOS:2 or 4, wherein the percent identity is determined using the ALIGN program of the GCG software package with a PAM120 weight residue table, a gap length penalty of 12, and a gap penalty of 4; and an amino acid sequence which is encoded by a nucleic acid molecule which hybridizes to the nucleic acid molecule consisting of any one of SEQ ID NOS:1, 3, 5, 6, 7, or a complement thereof, under conditions of hybridization of 6X SSC at 45°C and washing in 0.2 X SSC, 0.1% SDS at 65°C. After immunization, a sample is collected from the mammal that contains an antibody that specifically recognizes a HKNG1 polypeptide as exemplified in SEQ ID NOS:2 or 4, or portions thereof. Preferably, the polypeptide is recombinantly produced using a non-human host cell. Optionally, the antibodies can be further purified from the sample using techniques well known to those of skill in the art. The method can further comprise producing a monoclonal antibody-producing cell from the cells of the mammal. Optionally, antibodies are collected from the antibody-producing cell.

Please add the following paragraphs before the paragraph beginning on page 87, line 24:

C10

Under the terms of the Budapest Treaty on the International Recognition of the Deposit of Microorganisms for the Purpose of Patent Procedure, deposit of the following materials was made with the American Type Culture Collection (ATCC) of Rockville, MD, USA.

Applicants' assignees, Millennium Pharmaceuticals, Inc. and The Regents of the University of California, represent that the ATCC is a depository affording permanence of the deposit and ready accessibility thereto by the public if a patent is granted. All restrictions on the availability to the public of the material so deposited will be irrevocably removed upon the granting of a patent. The material will be available during the pendency of the patent application to one determined by the Commissioner to be entitled thereto under 37 C.F.R. 1.14 and 35 U.S.C. § 122. The deposited material will be maintained with all the care necessary to keep it viable and uncontaminated for a period of at least five years after the most recent request for the furnishing of a sample of the deposited plasmid, and in any case for a period of at least thirty (30) years after the date of deposit or for the enforceable life of the patent, whichever period is longer. Applicants' assignee acknowledges its duty to replace the deposit should the depository be unable to replace the deposit or should the depository be unable to furnish a sample when requested due to the condition of the deposit.

The clone of human DNA, Bluescript Epfsh 15w6, was deposited with the American Type Culture Collection (ATCC, Rockville, MD); received by the ATCC on March 6, 1997; and was assigned ATCC designation 98351.

In the claims:

Please amend claims 1, 19, and 21-31 as follows:

C11

1. (Amended TWICE) A method for identifying an individual having or at risk of developing a neuropsychiatric disorder comprising the step of detecting the presence or absence of *HKNG1* gene product in a patient sample wherein said method comprises the steps of:

a) incubating a sample in the presence of a detectably labeled antibody that identifies the *HKNG1* gene product; and